# POPULATION ECOLOGY OF THE FURBISH LOUSEWORT, *PEDICULARIS FURBISHIAE* S. WATS.

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The Furbish Lousewort is a rare and endangered plant species with a total of about 700 native individuals endemic to the St. John River Valley in Maine-New Brunswick. It was discovered by Kate Furbish in 1880 at Van Buren, Aroostook Co., Maine and described by Sereno Watson (1882) of Harvard University, where Furbish's original herbarium specimens of the species are deposited. A study by Macior (1978) indicated that *Pedicularis furbishiae* prefers calcareous, well-drained, sandy loam of north-facing, relatively stable but eventually transient, river terrace habitats shaded in part by a boreal coniferous forest. It is usually associated with Downy Alder (*Alnus crispa* (Ait.) Pursh), but mature plants of *Pedicularis furbishiae* were not observed to be root parasites as are some other species of *Pedicularis*. It reproduces by seed following pollination by bumblebee workers (*Bombus vagans* Sm.), which forage mostly for nectar but occasionally for pollen polylectically. Present reproductive rates of the plant in the field approximate replacement levels with some potential for population increase. Thus, the expansion of present populations appears immediately limited by availability of suitable habitat.

In view of the fact that *Pedicularis canadensis* and *P. lanceolata* of eastern temperate North America are root parasites (Piehl, 1963 & 1965) and since little is known about seedling development in the genus, the present study was designed to investigate seed germination, seedling development, and possible parasitic relationships in *Pedicularis furbishiae*.

## MATERIALS AND METHODS

Seeds of *Pedicularis furbishiae* were collected from populations at Allagash, Aroostook Co., Maine in the fall of 1977. One portion of seeds ("fall-sown") was surface sterilized for 10 minutes in 100 ml of a 10% commercial Clorox solution to which one or two drops of a liquid detergent were added. The seeds were rinsed several times with sterile water and incubated under sterile conditions on moist filter paper in 100mm × 20mm Petri dishes. The seeds were started on February 13–14, 1978. Cultures were maintained at room temperature under constant illumination. Since only 11 seeds germinated by March 23, the ungerminated seeds were transferred to sterile, moist sand in deep 100mm culture dishes.

A second portion of seed was given a cold treatment by exposure to four 10-day periods at 3° C alternating with four 10-day periods at 18° C, and was sown on March 23 on moist, sterile filter paper as with the fall-sown seed. Only 10 seeds germinated by Mar 31, at which time the remaining seeds were surface sterilized for 1 hr in a 1% Clorox solution, rinsed several times in sterile water, soaked for 24 hr in a 2,000 ppm solution of gibberellic acid ($A_3$), rinsed several times in sterile water and sown on moist, sterile filter paper in Petri dishes.

A third portion of seed was frozen from April 8 until May 23 and treated in the same manner as the cold-treated seeds.

Within 5 days the cold-treated seeds and the frozen seeds germinated abundantly. The fall-sown seed also began rapid germination on June 14. Seedlings were planted in a commercial "Terralite" potting mixture and grown in a controlled environment chamber under a 15 hr day at 400 ft candles and temperatures of 21° C during the day and 18° C at night.

In order to establish sterile cultures of *Pedicularis furbishiae* seedlings, small portions of fall-sown, cold-treated, and frozen seed treated as above were sown individually on sterile nutrient agar (Baslerova & Dvorakova, 1962) containing ammonium nitrate (0.2g), monobasic potassium phosphate (0.1g), magnesium sulfate (0.1g), calcium chloride (0.1g), ferric chloride (0.005g), dextrose (5.0g), and agar (10.0g) in 1 liter of distilled water.

The generally poor growth of *Pedicularis furbishiae* seedlings encountered under all experimental cultural conditions prompted a brief investigation of possible mineral nutrient deficiency. Potted seedlings were regularly nourished with a total of one liter of the following solutions: 0.1g/1 KCl (5 seedlings); 0.005g/1 $FeCl_3$ (6 seedlings); 0.2g/1 $NH_4NO_3$ (4 seedlings); 0.2 $NH_4NO_3$+0.1g $KH_2PO_4$+ 0.1g $MgSO_4$+0.1g $CaCl_2$+0.005g $FeCl_3$/liter (8 seedlings).

The possibility of obligate root parasitism in early stages of development of *Pedicularis* seedlings was investigated by sowing seeds of Crimson Clover (*Trifolium incarnatum* L.) with potted seedlings of *Pedicularis furbishiae* beginning on June 23. At this time all the experimental *Pedicularis* seedlings were small and chlorotic.

## RESULTS

The results on the germination of fall-sown, cold-treated, and frozen seeds are summarized in Table 1. Frozen and cold-treated seeds germinated at higher rates than fall-sown seeds. Since these higher rates of germination may have ben caused by treatment with gibberellic acid, however, the effects of thermal and hormonal treatment cannot be identified separately.

Seed germination in sterile cultures is shown in Table 2. Although all of the fall-sown seeds in sterile culture germinated, the small sample size may not be statistically significant for comparison with the results from the frozen and cold-treated seeds. Seedling growth was initially very rapid. The first true leaves appeared within 10 days of the protrusion of the radicle from the seed coat. In all cases subsequent seedling growth was very slow. The seedlings produced 4 to 6 small chlorotic leaves and suspended further growth. They did not, however, die immediately.

Table 1. **Seed Germination in *Pedicularis furbishiae*.**

| Treatment | Total Sown | Total Germinated | Percent Germination |
|---|---|---|---|
| Fall-sown | 1,045 | 322 | 31 |
| Frozen* | 603 | 541 | 90 |
| Cold-treated* | 636 | 561 | 88 |
| Total | 2,284 | 1,424 | 62 |

*These seeds were also treated with gibberellic acid ($A_3$) at 2,000ppm for 24 hr.

Table 2. **Seed Germination of *Pedicularis furbishiae* in Sterile Culture.**

| Treatment | Total Sown | Total Germination | Percent Germination |
|---|---|---|---|
| Fall-sown | 10 | 10 | 100 |
| Frozen* | 29 | 17 | 59 |
| Cold-treated* | 102 | 82 | 80 |
| Total | 141 | 109 | 77 |

*These seeds were also treated with gibberellic acid ($A_3$) at 2,000ppm for 24 hr.

The slow growth of potted seedlings supplied with supplemental nutrients may indicate that retarded growth was not caused by mineral deficiency. These seedlings remained small and chlorotic and eventually died.

The appearance of *Pedicularis furbishiae* seedlings grown with those of *Trifolium incarnatum* was in striking contrast to that of the controls without Crimson Clover (Fig. 1). The *Pedicularis* leaves expanded greatly, became dark green, proliferated abundantly, and were similar to those of young native plants in the field. This change occurred about 4 to 6 weeks after the *Trifolium* seedlings had become well established. Examinations of the roots of both plants clearly indicated the presence of *Pedicularis* haustoria firmly attached to the *Trifolium* roots. Morphologically and anatomically the haustoria closely resemble those of other *Pedicularis* species (Piehl, 1963 & 1965). The survival rates of *Pedicularis* seedlings with or without the *Trifolium* host are summarized in Table 3. Data were taken 20 weeks after *Trifolium* was sown with *Pedicularis*. The 53% loss of *Pedicularis* seedlings growing with *Trifolium* is in large measure due to the lack of development of haustorial attachments to the host. Eventually all the 97 seedlings that had survived without a host for 20 weeks died.

A preliminary test of wheat (*Triticum aestivum* L.) as a host for *Pedicularis furbishiae* roots gave similar results. Leaves of *Pedicularis* plants grown in association with wheat expanded and turned dark green, while those of the controls remained small and chlorotic.

In the fall of 1978 the older leaves on the surviving *Pedicularis furbishiae* seedlings began to die. Closer inspection revealed the formation of large terminal buds at the crown of each plant. Since these changes suggested the onset of dormancy, the environmental chamber temperature was lowered to a constant 15°C throughout the winter months. In the spring of 1979 the terminal buds expanded to produce short stems with pinnatifid leaves more closely resembling the cauline leaves of flowering plants than those of basal rosettes. Because of the difficulty of maintaining the annual *Trifolium* host with the perennial *Pedicularis furbishiae* seedlings, the latter eventually died.

## DISCUSSION AND CONCLUSIONS

The delay in germination of fall-sown *Pedicularis furbishiae* seeds

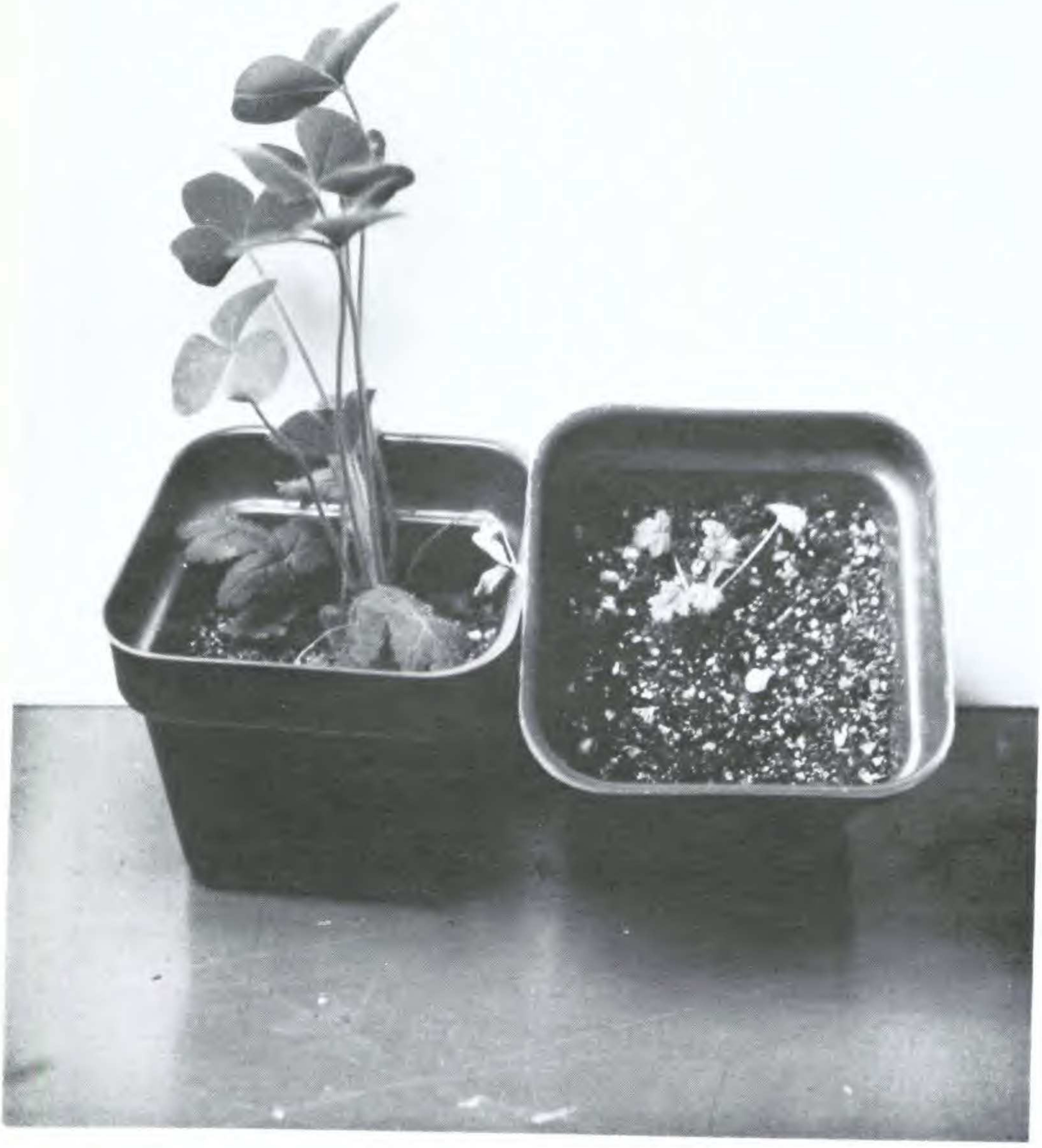

Fig. 1. Photograph taken on August 16, 1978 of *Pedicularis furbishiae* seedlings grown from frozen seed treated with gibberellic acid and planted June 8, 1978. Pot at left planted with *Trifolium incarnatum* June 30, 1978. Control without *trifolium* at right.ht. ×0.06.

Table 3. **Parasitic Survival of *Pedicularis furbishiae* Seedlings**

| Host | Total Sown | Total Surviving | Percent Survival |
|---|---|---|---|
| *Trifolium incarnatum* | 486 | 228 | 47 |
| None | 817 | 97 | 12 |

suggests that they may require an extended latent period coinciding with the dormancy period of plants in nature during the winter. Although the possible effect of the application of gibberellic acid to cold-treated and frozen seeds cannot be separated from the effects of low temperature *per se*, it is possible that thermal treatment hastens germination and that the application of gibberellic acid further enhances the process. That neither of these factors is absolutely necessary for germination is indicated by the substantial germination of untreated fall-sown seed.

The demonstration that *Pedicularis furbishiae* is an obligate root parasite in the seedling stage but apparently not obligately parasitic as an adult plant suggests that at early stages in development it requires some chemical component that it cannot synthesize or acquire from the physical environment directly. Since *Pedicularis* in nature grows in nitrogen-poor soil, *Trifolium* was chosen as an experimental symbiont for *Pedicularis* with the thought that nitrogen-fixing bacteria in the clover root nodules may provide a needed nitrogen supplement for seedling development. However, *Pedicularis furbishiae* did parasitize *Triticum* as effectively as *Trifoliium*, so this possibility can be eliminated. Furthermore, since neither of these cultivated host plants is native to the *Pedicularis* habitat, it is quite probable that *P. furbishiae*, like most other species known to be root-parasites (Sperlich, 1902; Maybrook, 1917; Sprague, 1962; Piehl, 1963 & 1965), is not host-specific. If this is so, it has an abundance of potential hosts in the diverse riparian plant community in which it grows.

The curious association of *Pedicularis furbishiae* in its native habitat with the perennial *Alnus crispa*, which is, in turn, obligately associated with an actinomycete on its roots, remains to be investigated. Seedlings of *Alnus* grown without the actinomycete closely resemble the small, chlorotic seedlings of *Pedicularis* without its host (Lalande, pers. comm.). Grown with the actinomycete, *Alnus* exhibits vigorous growth like that of *Pedicularis* growing with *Trifolium* or *Triticum*. The possible reciprocal or unilateral developmental relationship of seedlings of *Alnus crispa* and *Pedicularis furbishiae* in relation to root parasitism is currently under investigation.

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